IDAHO BUREAU OF LABORATORIES

IDAHO DEPARTMENT OF HEALTH & WELFARE

LABORATORY CONNECTIONS

Fall 2003

CHANGE

Richard Hudson, Ph.D. Bureau Chief

It is with a sense of sadness we acknowledge the dissolution of the branch laboratory system which for 60 years worked in partnership with the local health districts providing direct patient services. When changes in the occurrence of diseases such as the rise of STDs, natural disasters, epidemics, and food borne outbreaks occurred, the branch laboratory system responded by providing localized standardized testing. We salute the branch laboratory personnel for their dedication.

Over the years, the Idaho Bureau of Laboratories, IBL, has responded to the changing needs of the public health system, and we continue to do so. By centralizing testing and personnel, the IBL will be able to effectively address the challenges of newly emerging diseases, the dramatic increase in antibiotic resistance of microorganisms, as well as rising concerns regarding environmental contamination. In addition, we will have greater surge capacity in the event of an emergency. We will not be accessible in the same manner as before, but we will be able to provide more technical support to each of you. We realize a change of this magnitude may bring with it some problems. Please let us know when any arise.

This Issue

Respiratory Big Guns

Influenza

and

Pertussis

SARS UPDATE

The IBL can now perform testing for SARS associated Coronavirus by rt-PCR on respiratory specimens and total antibody serology on serum. Serology testing must include acute and 21-day convalescent sera. Contact the district health epidemiologist for submittal guidance.

A TRUE STORY

A routine drinking water sample tested positive for total coliform and *E. coli*. A swab taken from the differential medium was streaked to McConkey agar as part of a completed test. After 24 hours of incubation, two colony types were observed. The first, a typical coliform and the second, a small non-lactose fermenter. Both were transferred to a gram negative identification system. The results the next day were *Citrobacter freundii* and *Yersinia pestis*. Panic ensued.

To avoid this scenario:

- Know the limitations of the identification system you are using.
- Do a gram stain. *Y. pestis* is a bipolar 0.5 by 1.0-2 µm gram negative rod, found primarily in singles or pairs.
- Know characteristic growth patterns, ie. *Yersinia pestis* forms slow growing pinpoint (1-2mm) grey white to opaque colonies on sheep blood agar at 24 hours.

In a clinical laboratory, the scenario could be the isolation of a small gram-negative rod or coccobacillus from a sputum or blood culture. Do you have procedures and reagent in place to rapidly rule out *Y. pestis?* If you cannot rule out *Y. pestis* how would you report your results?

Influenza History Being Written

The CDC estimates influenza kills 36,000 in the U.S. each season. Influenza viruses undergo frequent antigenic changes, making vaccines obsolete and creating the very real threat of the emergence of a pandemic strain which could significantly increase infection and death rates.

Influenza viruses are tracked by a worldwide influenza surveillance system maintained by the World Health Organization. Last season, this surveillance detected human infections with two potential pandemic influenza strains; avian influenza A(H5N1) in Hong Kong and A(H7N7) in the Netherlands. The Idaho Bureau of Laboratories participates in this surveillance by culturing respiratory specimens, identifying and subtyping isolates. Representative isolates are forwarded to CDC for further antigenic studies. This information is used to detect antigenic shifts and to guide vaccine composition.

The availability of rapid testing for influenza viruses has greatly improved diagnostic capabilities in the clinical setting, but these tests do not provide the critical antigen shift information isolates provide. The sensitivity and specificity of the rapid tests is very good once influenza is widely circulating, but the probability of false positives greatly increases when prevalence is low. Positive rapid tests out of season should be confirmed by culture.

Year Round Surveillance!

In August an Influenza A (H3N2) was isolated from a patient who had traveled to China.

The trivalent Influenza vaccine for the 2003-04 season will include:

A/New Caledonia / 20/99-like (H1N1),

A/Moscow/10/99-like (H3N2), and

B/Hong Kong / 330/2001-like viruses.

Influenza Collection Kits / Testing FREE

The IBL maintains year-round influenza culture capability. We provide collection kits each season to a network of clinics and physicians in Idaho who help us with our viral surveillance efforts. Collection kits consist of a nasopharyngeal (NP) swab and transport media, a viral culturette for throat swab, a submittal form, and instructions for specimen collection.

Isolates are screened with a panel of monoclonal antibodies that will identify not only Influenza A and B, but also Parainfluenza 1, 2, 3, Respiratory Syncytial Virus, and Adenovirus. Because viral isolation can take up to two weeks for a negative result, we also provide a rapid Direct Fluorescent Antibody test on NP specimens.

We encourage anyone who would like to participate in the influenza surveillance to contact the Virology/Serology section at 208-334-2235.

Pieces of the Mosaic of *Bordetella pertussis* Testing

Collection

- Cotton swabs should never be used for collection of pertussis specimens.
- Nasopharyngeal swabs are better than throat swabs as *B. Pertussis* exhibits a tropism for ciliated respiratory epithelium found in the NP.

Recovery

- Best when done on site.
- Prior to starting antibiotics.
- Increased if specimen is taken within first two weeks.

Transport Affects Viability

- At room temperature, 55% of the organisms will die in the first 24 hours and 100% will be dead within 72 hours.
- If the Regan-Lowe transport is incubated for 1-2 days prior to transport to allow the organism to multiply, the transit time cannot exceed 3 days.
- When refrigerated, 75% of the organisms are not viable within the first 24 hours; however, the remaining 25% will remain viable for 8 days.

Submission of Respiratory Specimens for Viral Testing

Virus	Type of Specimen	Test	Result
Influenza A and B Parainfluenza 1, 2,3 Adenovirus Respiratory Syncytical Virus	NP swab in viral transport media. NP aspirate or wash. Throat swab in viral transport or viral culturette.	Culture	7-14 days
		DFA (on NP specimens only)	1 day
	Serum	IgM/IgG antibody by IFA	1-2 days
Mycoplasma pneumonia	Serum	IgM/IgG antibody by IFA	1-2 days
SARS-associated Coronavirus *	NP or OP swabs/washes/aspirates Broncheoalveolar lavage, Sputum	Real Time PCR	1-2 days
	Serum (Negative acute must be followed by 21-day convalescent specimen).	Total Antibody by EIA	1-2 days

NP refers to Nasopharyngeal, OP refers to oropharyngeal, * Prior consultation required.

Laboratory Diagnosis of Pertussis

Specimen	Test	Sensitivity	Specificity	Comments	Result
Slide from NP	DFA	LOW (When compared with culture)	Undetermined *	Detects organisms regardless of viability.	24 hrs
NP rayon or Dacron swab	Culture Gold Standard	MODERATE	HIGH	** Time of collection within disease process and transport critical.	10 days
NP rayon or Dacron swab	PCR	High Can detect non-viable cells ***	HIGH	Not FDA approved. Needs to be done in conjunction with culture and clinical case definition.	5 days

* False positives from cross-reactions with other organisms and false negatives due to low number of organisms.

** Negative culture results may be due to loss of viability rather than the absence of the organism.

*** Because non-viable cells are detected a positive result is not indicative of the presence of disease.

Editor's Corner

Sandra Radwin

Kari Getz shared the following information from a APHL workshopon Virology Methods.

- Because the "Flu Mist" is a live attenuated vaccine, it will grow nicely in cell culture making identification of actual influenza difficult.
- Influenza vaccine is now being recommended for children as this age group seems to spreads the virus most rapidly.
- Emerging viruses such as SARS have initially been identified by "traditional" methods such as cell culture rather then by the quicker methods such as PCR.

Call if you have any questions regarding specimen collection, packaging, or shipping.

For flu, SARS or RSV call:

208-334-2235 ext. 228

For pertussis, call:

208-334-2235 ext. 252

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"PROTECTING THE HEALTH AND ENVIRONMENT OF THE PEOPLE OF IDAHO THROUGH TESTING AND RESEARCH."

References: Overview of influenza and weekly updates about U.S. influenza activity can be seen at http://www.cdc.gov/ncidod/diseases/flu/fluinfo.htm

McGowan, K.L. 2002. Diagnostic Tests for Pertussis: Culture vs. DFA vs. PCR. Clin. Micro. *Newsletter.* **24:** 43-149.

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